

# PATENT COOPERATION TREATY

## PCT

### INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY


(Chapter II of the Patent Cooperation Treaty)

(PCT Article 36 and Rule 70)

REC'D 02 AUG 2005

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Applicant's or agent's file reference PPD70269/WO		<b>FOR FURTHER ACTION</b>		See Form PCT/PEA/416
International application No. PCT/GB2004/002447		International filing date (day/month/year) 09.06.2004		Priority date (day/month/year) 08.07.2003
International Patent Classification (IPC) or national classification and IPC C12N15/82, C12N9/06				
Applicant SYNGENTA LIMITED et al.				
<p>1. This report is the international preliminary examination report, established by this International Preliminary Examining Authority under Article 35 and transmitted to the applicant according to Article 36.</p> <p>2. This REPORT consists of a total of 6 sheets, including this cover sheet.</p> <p>3. This report is also accompanied by ANNEXES, comprising:</p> <p>a. <input type="checkbox"/> sent to the applicant and to the International Bureau) a total of sheets, as follows:</p> <p><input type="checkbox"/> sheets of the description, claims and/or drawings which have been amended and are the basis of this report and/or sheets containing rectifications authorized by this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions).</p> <p><input type="checkbox"/> sheets which supersede earlier sheets, but which this Authority considers contain an amendment that goes beyond the disclosure in the international application as filed, as indicated in item 4 of Box No. I and the Supplemental Box.</p> <p>b. <input type="checkbox"/> (sent to the International Bureau only) a total of (indicate type and number of electronic carrier(s)) , containing a sequence listing and/or tables related thereto, in computer readable form only, as indicated in the Supplemental Box Relating to Sequence Listing (see Section 802 of the Administrative Instructions).</p>				
<p>4. This report contains indications relating to the following items:</p> <p><input checked="" type="checkbox"/> Box No. I Basis of the opinion</p> <p><input type="checkbox"/> Box No. II Priority</p> <p><input type="checkbox"/> Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability</p> <p><input type="checkbox"/> Box No. IV Lack of unity of invention</p> <p><input checked="" type="checkbox"/> Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement</p> <p><input checked="" type="checkbox"/> Box No. VI Certain documents cited</p> <p><input type="checkbox"/> Box No. VII Certain defects in the international application</p> <p><input type="checkbox"/> Box No. VIII Certain observations on the international application</p>				
Date of submission of the demand  21.01.2005		Date of completion of this report  01.08.2005		
Name and mailing address of the international preliminary examining authority:  European Patent Office D-80298 Munich Tel. +49 89 2399 - 0 Tx: 523656 epmu d Fax: +49 89 2399 - 4465		Authorized Officer Morawetz, R Telephone No. +49 89 2399- 8155		



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**Box No. I Basis of the report.**

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1. With regard to the **language**, this report is based on the international application in the language in which it was filed, unless otherwise indicated under this item.
- ☐ This report is based on translations from the original language into the following language , which is the language of a translation furnished for the purposes of:
- ☐ international search (under Rules 12.3 and 23.1(b))
  - ☐ publication of the international application (under Rule 12.4)
  - ☐ international preliminary examination (under Rules 55.2 and/or 55.3)
2. With regard to the **elements\*** of the international application, this report is based on *(replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report)*:

**Description, Pages**

1-52 as originally filed

**Sequence listings part of the description, Pages**

1-3 as originally filed

**Claims, Numbers**

1-17 as originally filed

**Drawings, Sheets**

1/1 as originally filed

☒ a sequence listing and/or any related table(s) - see Supplemental Box Relating to Sequence Listing

3. ☐ The amendments have resulted in the cancellation of:
- ☐ the description, pages
  - ☐ the claims, Nos.
  - ☐ the drawings, sheets/figs
  - ☐ the sequence listing (*specify*):
  - ☐ any table(s) related to sequence listing (*specify*):
4. ☐ This report has been established as if (some of) the amendments annexed to this report and listed below had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).
- ☐ the description, pages
  - ☐ the claims, Nos.
  - ☐ the drawings, sheets/figs
  - ☐ the sequence listing (*specify*):
  - ☐ any table(s) related to sequence listing (*specify*):

\* If item 4 applies, some or all of these sheets may be marked "superseded."

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**Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement**

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**1. Statement**

Novelty (N)	Yes: Claims	1-17
	No: Claims	
Inventive step (IS)	Yes: Claims	1-17
	No: Claims	
Industrial applicability (IA)	Yes: Claims	1-17
	No: Claims	

**2. Citations and explanations (Rule 70.7):**

**see separate sheet**

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**Box No. VI Certain documents cited**

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**1. Certain published documents (Rule 70.10)**

and /or

**2. Non-written disclosures (Rule 70.9)**

**see separate sheet**

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**Supplemental Box relating to Sequence Listing**

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**Continuation of Box I, item 2:**

1. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application and necessary to the claimed invention, this report has been established on the basis of:
  - a. type of material:
    - ☒ a sequence listing
    - ☐ table(s) related to the sequence listing
  - b. format of material:
    - ☒ in written format
    - ☒ in computer readable form
  - c. time of filing/furnishing:
    - ☒ contained in the international application as filed
    - ☒ filed together with the international application in computer readable form
    - ☐ furnished subsequently to this Authority for the purposes of search and/or examination
    - ☐ received by this Authority as an amendment on
2. ☐ In addition, in the case that more than one version or copy of a sequence listing and/or table(s) relating thereto has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.
3. Additional observations, if necessary:

**Re Item V**

**Reasoned statement with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement**

1. The following documents are referred to in this communication:

- D1 : WO 98/39462 A (CIBA GEIGY AG ; CROSSLAND LYLE DEAN (US); PASCAL ERICA (US); HARPER ST) 11 September 1998 (1998-09-11)
- D2 : WO 02/072804 A (BAYER CROPSCIENCE N V ; PUEHLER ALFRED (DE)) 19 September 2002 (2002-09-19)
- D3 : WO 01/29237 A (AVENTIS CROPSCIENCE N V) 26 April 2001 (2001-04-26)
- D4 : PILONE M S: "D-amino acid oxidase: new findings" CMLS CELLULAR AND MOLECULAR LIFE SCIENCES, BIRKHAUSER VERLAG, BASEL, CH, vol. 57, 2000, pages 1732-1747, XP002967442 ISSN: 1420-682X
- D5 : SACCHI SILVIA ET AL: "Engineering the substrate specificity of D-amino acid oxidase" JOURNAL OF BIOLOGICAL CHEMISTRY, vol. 277, no. 30, 28 July 2002 (2002-07-28), pages 27510-27516, XP002295815 ISSN: 0021-9258

2. Subject-matter of the application

Present application relates to methods of producing male and female sterile plants using a non-phytotoxic substance and expressing in a tissue-specific manner an enzyme which converts the non-phytotoxic substance into a phytotoxic one. In particular, the non-phytotoxic substance is a D-alpha amino acid, or a peptide derivative of a non-protein D-alpha amino acid, the D enantiomer of phosphinothricin or a D enantiomer of bialaphos, and the enzyme is a mutant D-amino acid oxidase obtainable from *Rhodotorula gracilis* which comprises a lysine at position 58 rather than a phenylalanine in the wild type sequence.

3. The subject-matter of claims 1-17 appears to be novel and inventive in view of the available prior art.
- 3.1. Document D1, which is considered to represent the most relevant state of the art to the subject matter of claim 1, discloses (page 8, third full paragraph - page 9,

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(SEPARATE SHEET)**

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second paragraph; page 17, line 3- line 27) various methods for the production of conditional female or male sterility in plants. According to D1 (page 9, second paragraph) any coding sequence which encodes an enzyme which catalyses the conversion of a protoxin to a toxin may be used. Examples disclosed in D1 are N-acetyl-L-ornithine deacetylase (argE) which converts the protoxin N-acetyl phosphinothricin (N-acetyl-PPT) to the toxin phosphinothricin (PPT); P450<sub>su1</sub> monooxygenase which converts the sulfonamide protoxin and phosphonate monoester hydrolase (pehA) which converts the protoxin glyceryl glyphosate. The subject-matter of independent claim 1 differs from the disclosure of D1 in that the enzyme is a mutant D-amino acid oxidase obtainable from *Rhodotorula gracilis* which comprises a lysine at position 58 rather than a phenylalanine in the wild type sequence and the protoxin is a D-alpha amino acid.

Since neither the specific protoxin nor the enzyme have been disclosed or suggested in the available prior art for the claimed use novelty and inventive step can be acknowledged.

- 3.2. Claim 16 which relates to a mutant D-amino acid oxidase obtainable from *Rhodotorula gracilis*, capable of oxidising phosphinothricin, which comprises a lysine at position 58 rather than a phenylalanine in the wild type sequence is likewise considered novel and inventive.
- 3.3. Claims 2-15 and 17 are dependent on claim 1 and 16, respectively, and as such also meet the requirements of the PCT with respect to novelty and inventive step.

**Re Item VI**

**Certain documents cited**

**Certain published documents**

Application No Patent No	Publication date (day/month/year)	Filing date (day/month/year)	Priority date (valid claim) (day/month/year)
PCT/EP03/00222	24.07.2003	13.01.2003	17.01.2002
PCT/GB03/00683	04.09.2003	14.02.2003	26.02.2002